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Richmond, Virginia

To: Charles Glenn

Date: June 30, 1992

From:

Debbie Ayers, Marc Krauss, Maria Shulleeta, and Samir Wahab

Subject: Plans to Complete Work in Support of PM Patent Application 1412

The following is a list of proposed research that we plan to do in support of PM patent application 1412. We are interested in determining, from a legal point of view, what is most important for us to accomplish during the next several months. We are therefore requesting your assistance. Please review these plans and communicate to us your suggestions regarding these plans and any additional work that should be done.

- 1. Transform tobacco (Burley 21) with sense and anti-sense constructs of the PMT gene. These plants will be sampled to determine alkaloid levels before and after topping. Seed will be collected. If time permits, heritability of any reduced alkaloid levels will be determined.
- 2. Isolate and sequence the entire PMT gene. This intact gene will be cloned into an expression vector and tested for expression (initially in \underline{E} . coli).
- 3. Antibodies generated to the 60 kD fragment (putative PMT) and another set of antibodies generated from a synthetic peptide (29 amino acids at the N-terminus) will be used to develop an affinity column and to screen the cDNA expression library.

cc: R. Carchman

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